359 Negative genetic neonatal screening for cystic fibrosis caused by compound heterozygosity for two large CFTR rearrangements

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Newborn screening programs for CF are applied in France for almost 800,000 newborns each year, based on IRT assay from dried blood spots, followed, when IRT is positive, by testing for the 30 most common CFTR mutations. Infants with positive sweat test are further investigated for CFTR point mutations, which usually results in the identification of two mutant alleles. We experienced a case of CF with meconium ileus positive but with no mutation after sequencing of CFTR coding/flanking regions. By using a semi-quantitative fluorescent PCR assay, we detected a deletion of exons 2, 18, 19, 20 previously named 2, 16, 17a, 17b. Segregation analysis and extensive study of the CFTR locus revealed that the neonate carried the previously described [c.54–5811_c.164+2186del8108ins182], inherited from his father, and a novel [c.2908+1085_c.3367+260del7201] rearrangement, inherited from his South Korean mother, involving exons previously reported in another deletion [c.2909–636_c.3368+1611del6965ins32]. In our knowledge, this is the first case of compound heterozygosity for two CFTR large rearrangements. This study emphazises on several points: (1) CF can be found in virtually every ethnic group, including non-Caucasian Asiatic populations; (2) CF can be caused by the inheritance of two heterozygous large rearrangements; (3) The same exons can be involved in different rearrangements; (4) Using the original CFTR gene data rather than the international nomenclature for gene exons and mutations may lead to misdiagnosis; (5) Sequencing the breakpoint junction is recommended because misdiagnosis could result from the use of rapid PCR assays designed to only test junctions of rearrangements.

360 Pulmonary and nutritional outcomes in children with cystic fibrosis (CF) detected through neonatal screening and by symptoms in Argentina

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Screening for CF in the newborn period has been established in many regions of the world. Previous studies have shown that the early treatment in the newborn period results in improved nutritional and clinical outcomes.
Objective: To compare pulmonary and nutritional status in children with CF detected through neonatal screening and by symptoms in follow-up at our center.
Methods: Cross sectional study. We assessed two groups of patients matched by age, sex and genotype; a group A (GA) detected by neonatal screening, and a group B (GB) by symptoms; both diagnosed by sweat test. Immunoreactive trypsinogen analysis (IRT) was used for neonatal screening (cut-off value 70 ug/ml). We evaluated age at diagnosis, age of first isolation of *Pseudomonas aeruginosa* (Pa), number of hospital admissions (A). Also we assessed Z score for weight/age (W/A) at first visit in our center, and Z score of Body Mass Index (BMI), Z score for height/age (H/A), score Shwachman Brasfield (S/B) and FEV1 at last visit. Chi2 and Wilcoxon tests were performed to compare the results.
Results: 21 children with CF were enrolled for each group (12 boys and 9 girls). Eleven patients for each group had mutations class II in both alleles.
Conclusions: Early diagnosis and therapeutic management of CF detected through neonatal screening result in significantly enhanced in nutritional and pulmonary status. In developing countries the carrying out of screening programs must be considered.

	n	X age (years)	Age at diagnosis (years)*	z W/A*	z BMI	z H/A*	S/B*	FEV1 (%)	A*	Age 1st Pa
GA	21	4.2	0.16	–0.66	0.19	0.1	81.2	99	1.42	1.11
GB	21	4.4	1.03	–2.23	0.17	–0.68	72.7	81	3.28	1.67

*$p < 0.05$

361 Pulmonary function in infants with cystic fibrosis (CF) detected through neonatal screening and by symptoms in Argentina

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Rationale: The most of the patients with Cystic Fibrosis (CF) is detected through neonatal screening or by symptoms. Lung disease in CF is at the beginning a disease mainly of the small airways. Although infants with CF have normal lung at birth, respiratory dysfunction often develops in the first year.
Objective: To compare pulmonary function in children younger than 24 months detected through neonatal screening and by symptoms in follow-up at our center.
Methods: Cross sectional study. We measured: Functional Residual Capacity (FRC), Airway Resistance (Raw), and Maximal Expiratory Flow at FRC (V'maxFRC) with Jaeger® equipment. We compared two groups of patients with similar age: group A detected by neonatal screening (GA), and group B by symptoms (GB), both diagnosed by sweat test. For neonatal screening we used immunoreactive trypsinogen (IRT) in dried blood spots (cut-off value: 70 ug/ml). Student's t-test was performed to compare the means of the indicators using the SSPS software 9.0.
Results: 20 infants with CF were enrolled (8 in GA and 12 in GB) with a mean age of 15.9±8 months, height 73±9 cm, z score for weight/age –1.2 and height/age –1.3. Mean V'maxFRC was 173.4±96 ml/s and its z score –1.34±1.6. FRC was 263.9±135 ml and Raw 1.67±1.2. Fourteen patients finished all measurements (6 GA and 8 GB).
Conclusion: Our infants with Cystic Fibrosis detected through neonatal screening had higher V'maxFRC and lower FRC and Raw than non-screened group. These findings should be taking into account when CF programs are considered.

	n	Height (cm)	V'maxFRC (ml/s*	z V'maxFRC*	FRC (ml)	Raw
GA	6	68.1	229	–0.06	191	0.71
GB	8	69.7	104	–2.19	224	1.70

*$p < 0.05$

362 Analytical characteristics of the Dynabio® pancreatitis associated protein (PAP) enzyme linked immunosorbent assay (ELISA)

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Aim: To evaluate the analytical characteristics of the Dynabio® PAP ELISA.
Methods: Analytical precision, linearity, lower limit of detection and potential interferences were determined according to Clinical & Laboratory Standards Institute guidelines EP5-A, EP6-A, EP7-A, EP9-A2 using recombinant human PAP (rhPAP) measured with the Dynabio® PAP ELISA (Marseille, France) according to manufacturer's specifications.
Results: Within-run precisions were 0.9% and 11.8% at 0.25 and 0.015 μg/L concentrations of rhPAP, respectively. The lower limit of detection was 0.015μg/L. The reportable range was 0.03 to 0.125 μg/L when the data were analyzed as optical density (OD) versus concentration. A double reciprocal plot of optical density versus rhPAP concentration extended the assay linearity (0.015 to 0.25 μg/L). The equations of the line and correlation coefficients were $y = 7.7(x) + 0.220$ and $r = 0.985$; $y = 0.07(x) + 0.123$ and $r = 0.994$ for OD versus concentration and 1/OD and 1/concentration analyses, respectively.
Conclusions: 1) The precision and lower limit of detection of the Dynabio® PAP ELISA (Marseille, France) meet analytical expectations for newborn cystic fibrosis screening. 2) The reportable range of linear results is extended when double reciprocal graphing of optical density versus rhPAP concentration is conducted.